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of the *C. obsoletus* complex, *C. chiopterus*, *C. dewulfi* and *C. punctatus* was 0.7, 0.4, 0.5 and 7.8 midges/minute, respectively. The overall biting rate (BR) was however low, with only 4.6% of the more than 5,000 midges captured off the horses found freshly blood-engorged. Both *C. chiopterus* and *C. dewulfi* were reared from the dung of the two experimental horses.

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Mosquito species presence on equine premises in the UK

G.E. Chapman, D.C. Archer, M. Baylis
Department of Epidemiology and Population Health, Institute of Infection and Global Health, University of Liverpool, UK

Background: Globally there has been increasing concern over emerging infectious diseases. In particular arboviral diseases represent a significant threat to animal and human health and the introduction of West Nile virus into North America has demonstrated the potentially significant effects of these viruses on a naïve host population. In Europe recent arboviral emergence events include Usustu, chikingunya, and dengue and in the UK, bluetongue and Schmallenberg viruses. A number of arboviruses affecting horses have been noted as important zoonoses with potential for emergence in Europe, including Japanese encephalitis virus, Eastern equine encephalitis virus, Western equine encephalitis virus and Venezuelan equine encephalitis virus. West Nile virus already causes outbreaks in equines in Europe including in Northern Italy and Southern France in 2015. Information regarding the species presence and numbers of mosquitoes on equine premises in the UK are lacking. This information is important in assessing the future risk to horses from mosquito borne disease. The purpose of this study was to add to information on mosquito presence and abundance on equine premises and to investigate the practicalities of mosquito surveillance techniques on equine premises in the UK. Initial fieldwork has concentrated on the species and relative numbers of mosquitoes present on equine premises and to provide baseline information for future mosquito surveillance. This study involved mosquito sampling on 8 premises in each of 4 widely distributed regions of England. Stratified sampling of four mosquito habitats in proximity to equine premises was utilised: Fenland, Woodland, Urban, and Saltmarsh. Each region was visited 3 times during the mosquito season and the timing of these visits was based around the average peaks of different mosquito species abundance. The main trap used for sampling was the Mosquito Magnet ™ and this was run for approximately 72 hours at each visit. Larval sampling and a resting box trap were also used on all sites, and resting mosquito sampling, and sweep netting were also used where appropriate. Mosquitoes were identified as far as possible morphologically and by molecular methods for the *Culex pipiens* complex. Preliminary results indicate that species presence is highly correlated with expected presence based on proximity to mosquito breeding habitat. *Cx pipiens* complex mosquitoes and *Culiseta annulata* were two of the most commonly found species. Further conclusions may be drawn after completion of the study and analysis of data.

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Long-lasting non-primate hepacivirus infection and transmission of the virus from dams to infants in horses

T. Miyashita[1], Y. Toishi[2], M. Hashimoto[2], N. Tsunoda[2], R. Kirisawa*[1]
[1] *Laboratory of Veterinary Virology, Department of Pathobiology, School of Veterinary Medicine, Rakuno Gakuen University, 582 Bunkyoudai Midori-machi, Ebetsu, Hokkaido 069-8501, Japan;* [2] *Shadai Corporation, 275 Hayakitagenbu, Abira, Hokkaido 059-1432, Japan*

Non-primate hepaciviruses (NPHVs) that infect horses are the closest relatives of hepatitis C virus (HCV) described to date. HCV and NPHV belong to the genus *Hepacivirus*, within the family *Flaviviridae* of single-stranded, positive sense RNA viruses. In this study, we analyzed the NPHV prevalence in stallions, broodmares and yearlings in Japan. We used 35 serum samples from stallions at Farm A and 122 and 125 serum samples from broodmares and yearlings at Farm B and Farm C, respectively, obtained in November 2013. All of the horses were Thoroughbred. Most of the yearlings were reared at Fam B with their dams until 6 to 7 months of age, and then moved to Farm C. NPHV RNA in serum samples was detected by a nested RT-PCR targeting conserved sequences in the nonstructural 3 (NS3) protein coding region. Four (11.4%) of the 35 stallions, 7 (5.8%) of the 122 broodmares and 4 (3.2%) of the 125 yearlings were NPHV RNA-positive. The NS3 region sequences of all 15 NPHV RNA-positive PCR products were determined. Phylogenetic analysis showed that all of the 15 NS3 sequences clustered with sequences previously classified as NPHV. The analysis revealed that vertical transmission from one broodmare to her infant and horizontal transmission among yearlings occurred. A retrospective and follow-up survey of NPHV RNA-positive horses revealed that most of the horses had chronic infection and that only a few horse had acute infection. Two horses showed viremia over a ten-year period. In 4 NPHV RNA-positive stallions, GGT, AST and LDH levels in serum were almost all within reference ranges. Therefore, hepatitis was not observed. This study suggested that NPHV infections are widespread in Japanese horses as well as in horses in other countries, such as UK, Germany, USA and Brazil, and that both vertical transmission and horizontal transmission occur.

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Genetic and antigenic analysis of Getah virus isolated in 1978 and 2014 in Japan

M. Nemoto*, H. Bannai, K. Tsujimura, T. Yamanaka, T. Kondo
Epizootic Research Center, Equine Research Institute, Japan Racing Association, Shimotsuke, Tochigi, Japan

Getah virus is a mosquito-borne virus that causes fever, rash on the body, and edema of the legs in horses and pigs. An inactivated vaccine has been used since the first outbreak occurred in 1978 at the Miho training center of the Japan Racing Association, and no outbreaks had occurred in vaccinated horses until 2014, when the disease resurfaced among racehorses at the same training center in September and October of that year [1]. As we reported previously [2], indirect causes of this outbreak likely include the existence of susceptible horses that have not completed the vaccination program and an increased risk of exposure to the virus because of epizootic Getah virus infection around the training center. However, the direct cause of the 2014 outbreak remains unclear. To determine whether mutation of the virus was directly responsible for the outbreak, we performed genetic and antigenic analysis of the vaccine strain (MI-110) isolated in 1978 and a strain isolated in 2014 (14-I-605). We sequenced the complete genomes of two cloned MI-110 strains and two cloned 14-I-605 strains. In addition, the original MI-110 and 14-I-605 were inoculated into seronegative horses. Antisera collected from these horses were used in a cross-neutralizing test. The complete genome sequences of the cloned 14-I-605 strains had 98.6% nucleotide identity to those of the cloned MI-110 strains. The nucleotide lengths of non-structural polyprotein (nsP1234, 7404

base pairs) and structural polyprotein (C-E3-E2-6K-E1, 3762 base pairs) were identical between MI-110 and 14-I-605. The nucleotide and amino acid sequences of these genes were highly conserved between MI-110 and 14-I-605 (nsP1234, 98.5% for nucleotides and 99.5% for amino acids; C-E3-E2-6K-E1, 98.7% for nucleotides and 99.8% to 99.9% for amino acids). Antisera against MI-110 and 14-I-605 had almost equal titers against both the homologous virus and the heterologous virus. These results showed that there was little difference between MI-110 and 14-I-605 genetically and antigenically. Therefore, the current vaccine containing MI-110 should be effective against 14-I-605, and it is unlikely that virus mutation was a direct cause of the 2014 outbreak.

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Identification of equine hepacivirus infections in France: Facts and Physiopathological insights

S. Pronost*[1,2,3,4], E. Hue[1,2,3,4], C. Fortier[1,4], M. Foursin[5], G. Fortier[1,2,3,4], F. Desbrosse[6], F. Rey[7], P.-H. Pitel[1,4], B. Saunier[7]
[1] *LABÉO Frank Duncombe, 14053 Caen, France;* [2] *Normandie Université, 14000 Caen, France;* [3] *Unité Recherche Risques Microbiens (U2RM), EA 4655, 14032 Caen, France;* [4] *Hippolia Foundation, 14000 Caen, France;* [5] *Clinique Equine de la Boisrie, 61 Chailloué, France;* [6] *Clinique Equine Desbrosse, 78 Saint Lambert, France;* [7] *Structural Virology Unit - CNRS UMR 3569, Institut Pasteur, 75 Paris, France*

Hepatitis C virus (HCV) is one of the main causes of end stage liver diseases (cirrhosis and hepatocellular carcinoma) in humans. *In vivo* studies have been hampered by a lack of access to relevant animal models. Until recently, the genus *Hepacivirus* was comprised of seven HCV genotypes, plus GBV-B that infects New World monkeys. Within the past few years, new hepaciviruses were identified in dogs and horses (also called canine/equine or non-primate hepaciviruses), in Old World monkeys (*Colobus guereza*), as well as in rodents (*Myodes glareolus* and *Rhabdomys pumilio*) and bats. Yet, the data available regarding newly identified hepacivirus infections is still limited, including in horses. So far, few studies have reported PCR positive horses in different countries (USA, Scotland, Germany, Japan, Brazil and Hungary). The aim of our study was to determine the prevalence of active hepacivirus infections in horses. A RT-qPCR adapted from the test described by Burbello et al. was developed according to the AFNOR norm U47-600-2. We detected 69 positive horses out of 1229 screened with this assay (5,6%). Positive horse samples originated from all parts of France. The number of viral genomes circulating in the blood of hepacivirus-infected horses ranged from 1.15 x 10^4 to 2.8 x 10^9 copies/mL, with two third of the results above a threshold of 4.26 x 10^7 copies/mL. There was no evidence of concomitant hepatic inflammation in the sera, as tested by the levels of gamma-glutamyl transferase and glutamate deshydrogenase. A one-year follow-up study in 7 horses from the same stud farm showed that, from the time of detection, infection persisted up to 18 months. A phylogenetic analysis was performed based on the nucleotide sequences of the 5'-untranslated region of the genome, as well as of the genes encoding the nonstructural proteins 3 and 5B. The result of this analysis suggests that all positive horses identified in France are infected with equine hepacivirus variants distinct from those previously reported. The present study demonstrates for the first time the presence of a significant proportion of ongoing equine hepacivirus infections in the horse herds in France. Determining the consequences of persistent hepacivirus infections in horses warrants further investigation.